Pathogenicity of Trypanosoma brucei in experimentally infected pigs

MATERIALS AND METHODS

Experimental animals
Six female Large White pigs, 4 to 5 months of age, were obtained from a farm near Ibadan. They were dewormed with piperazine citrate at a dosage of 400 mg per kg live-weight, screened for blood parasites and confirmed to be free of trypanosomosis. The pigs were housed in a fly-proof accommodation and fed a commercial growers ration (Pfizer Livestock Feeds, Ibadan, Nigeria) throughout the study period. Male Albino mice were obtained from the experimental Animal Unit of the College of Medicine, University of Ibadan.

Trypanosoma brucei stock
The stock was obtained from an anaemic local pig at slaughter at the Ibadan Municipal Government abattoir, with a parasitaemia of $10^3$ trypanosomes per ml blood.

After staining with Giemsa, the parasites were identified as a T. brucei species. The organisms were infective for mice. When subjected to the blood incubation infectivity test (10), infectivity of the parasites for mice was abolished by the action of human serum, but not bovine or porcine serum. The parasites were thus identified as Trypanosoma b. brucei.

Experimental protocol
Four of the 6 experimental pigs were inoculated intravenously with mouse blood containing approximately $5 \times 10^6$ motile T. brucei parasites. The remaining two pigs served as non-infected controls.

Every other morning, between 9 and 10 a.m., the body temperature was measured with a rectal thermometer and blood was collected from an ear vein into two heparinized centrifuge tubes for determination of packed cell volume and parasitaemia, as described by HERBERT and LUMSDEN (2).

All pigs were euthanised at the end of observation, 50 days post infection (dpi). Tissues for histopathology were fixed in 10% buffered formalin, processed routinely for histopathology and stained with haematoxylin and eosin. Selected sections were stained with Giemsa or Martin.
Scarlet blue. Wet mounts were made from the cerebrospinal fluid (CSF) and examined for trypanosomes. Smears made from CSF were also stained with Giemsa and examined for trypanosomes.

RESULTS

Clinical findings and haematology

The mean prepatent period of parasitaemia was 4.5 ± 0.6 (SD) days. Subsequently, there was undulating parasitaemia in infected pigs with peaks occurring at intervals of 5-8 days in each animal. Peak parasitaemia in individual infected pigs varied from $10^7$ to $10^4$ trypanosomes per ml blood (fig. 1).

A rise in the mean rectal temperature of infected pigs became apparent at 4 dpi (fig. 2). Thereafter, the mean rectal temperature of infected pigs remained higher than that of the controls even though the values fell to normal levels at intervals in individual pigs. The peak of rectal temperature in individual pigs varied from 41.1 to 41.8 °C.

By 50 dpi, the mean packed cell volume had fallen by 34 % i.e. from 40 to 26 % (fig. 1). Infected pigs were anorectic, weak, recumbent and very lean. A pig started showing intermittent circling and wobbling of the hindlegs as from 46 dpi.

Pathology

At necrospy, the carcasses of infected pigs lacked subcutaneous or abdominal fat and were lean. The lymph nodes especially the prescapular, subcutaneous and those of the lumbar region, were markedly enlarged. The brain of the pig that showed nervous signs was grossly congested and focal grey areas of necrosis were present on the cerebral cortex.

Microscopical examination of tissues of all infected pigs revealed moderate haemosiderosis, erythrophagocytosis, neutrophilic and eosinophilic infiltration of the white pulp as well as a mild macrophage and plasma cell hyperplasia in the spleen. The ovarian lesion was characterized by mild eosinophilic, lymphocytic and macrophage infiltrates into the connective tissue. In addition, there was sludging of lymphocytes and neutrophils in
many blood vessels. In all infected pigs a marked congestion and oedema was observed in the lymph nodes, with medular and subcapsular spaces containing moderate neutrophilic infiltrates. A moderate macrophage hyperplasia was also seen in the medular area of the lymph nodes. In the pig that exhibited nervous signs, the brain lesion was a very severe meningoencephalitis which was characterized by a very severe lymphocytic infiltrates into the meninges, vasculitis, perivascular cuffs (photo 1) and glia nodules in the brain tissue. The presence of trypanosome nuclei both in the brain tissue and in the blood vessels was confirmed in Giemsa stained sections. Motile trypanosomes were seen in the wet mounts made from CSF while the Giemsa stained smears also contained trypanosomes. Similar lesions were not seen in the brain of the remaining three infected pigs and no lesions were observed in the control pigs.

DISCUSSION

The observations on parasitaemia, rectal temperature and packed cell volume in this study are consistent with those reported for virulent species of African trypanosomes (11). The results also indicate that the stock of *Trypanosoma brucei* used in this study could cause economic losses through anaemia and invasion of the central nervous system which invariably terminates fatally (3, 4, 5).

The meningoencephalitis observed at histopathology and the presence of trypanosomes in the CSF and brain tissue explains the signs of central nervous system disturbance observed in one of the pigs. This observation has not been previously shown in pigs infected with *T. brucei* although meningoencephalitis has been observed in the dog and horse naturally infected with *T. brucei* (3, 4, 5). VOHRADSKY (13) and ISOUN and ANOSA (6) reported the presence of endometritis and cystic ovaries in cattle and sheep infected with *T. vivax*. The lesions were not observed in pigs infected with *T. brucei* in the present study although eosinophilic and macrophage infiltrates were observed in the ovarian connective tissue of infected pigs.

The present results thus support a previous finding in Northern Nigeria that *T. brucei* may cause a severe rather than a mild syndrome in infected pigs (1).
REFERENCES


